309. *The Action of Alcoholic Monomethylamine on Derivatives of Benzoquinone and Toluquinone. Part I. The Methoxy- and Hydroxymethoxy-derivatives.*

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An investigation has been made of the action of alcoholic monomethylamine on (A) the mono- and di-methoxy-derivatives of benzoquinone and on the mono-, di-, and tri-methoxy-derivatives of toluquinone and (B) a number of hydroxymethoxy-derivatives of benzo- and tolu-quinone. (A) With two exceptions the reaction proceeded in the anticipated way. 4-Methoxytoluquinone and 4 : 6-dimethoxytoluquinone behaved abnormally, giving respectively 2 : 5-*bismethylamino*-1 : 4-*benzoquinone* and 2 : 5-*bismethylamino*-3-*methoxy*-1 : 4-*benzoquinone*, which were identified by their hydrolysis products, 2 : 5-dihydroxybenzoquinone and 2 : 5-dihydroxy-3-methoxybenzoquinone. This abnormal reaction was not observed with any other methoxy-derivative having a methoxyl group *para* to methyl. (B) Certain other abnormalities were observed and the methylamino-derivatives obtained and also their hydrolytic products are described. Methods are given for the preparation of 3 : 4 : 6-*trimethoxytoluquinone*, 2-*hydroxy*-5-*methoxybenzoquinone*, 5-*hydroxy*-2 : 3-*dimethoxybenzoquinone*, 6-*hydroxy*-3-*methoxytoluquinone*, 6-*hydroxy*-3 : 4-*dimethoxytoluquinone* and their respective quinols 2 : 5-*dihydroxy*-3 : 4 : 6-*trimethoxytoluene*, 1 : 2 : 4-*trihydroxy*-5-*methoxybenzene*, 1 : 4 : 5-*trihydroxy*-2 : 3-*dimethoxybenzene*, 2 : 5 : 6-*trihydroxy*-3-*methoxytoluene* and 2 : 5 : 6-*trihydroxy*-3 : 4-*dimethoxytoluene*.

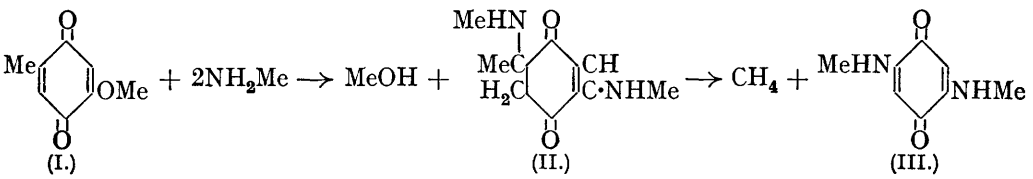
Spinulosin, a metabolic product of the mould *Penicillium spinulosum* Thom (Birkinshaw and Raistrick, *Phil. Trans.*, 1931, *B*, **220**, 245), was shown by Anslow and Raistrick (*Biochem. J.*, 1938, **32**, 687, 803) to be 3 : 6-dihydroxy-4-methoxy-2 : 5-toluquinone. Applying the well-known method for the synthesis of *p*-dihydroxyquinones, an attempt was made to synthesise spinulosin by the interaction of alcoholic monomethylamine and 4-methoxytoluquinone and hydrolysis of the resulting methylamino-derivative. Instead of the expected 3 : 6-bismethylamino-4-methoxytoluquinone, however, 2 : 5-*bismethylamino*-1 : 4-*benzoquinone* was formed, which gave 2 : 5-dihydroxy-1 : 4-benzoquinone on hydrolysis. This surprising reaction led us to investigate the action of alcoholic monomethylamine on substituted benzo- and tolu-quinones and the present communication records the results obtained with a number of methoxy- and hydroxymethoxy-derivatives of benzo- and tolu-quinone.

Benzoquinone, methoxybenzoquinone, and 2 : 5-dimethoxybenzoquinone reacted in the expected way and gave 2 : 5-*bismethylamino*-1 : 4-*benzoquinone*. Similarly, toluquinone, as was already known (Fichter, *Annalen*, 1908, **361**, 400), 3-methoxytoluquinone, 6-methoxytoluquinone and 3 : 6-dimethoxytoluquinone reacted normally and gave 3 : 6-bismethylamino-2 : 5-toluquinone, which gave 3 : 6-dihydroxytoluquinone on hydrolysis. On the

other hand, 4 : 6-dimethoxytoluquinone reacted in a similar way to 4-methoxytoluquinone, losing the methyl group and the methoxy-group in the *para*-position to it, and gave 2 : 5-*bismethylamino*-3-*methoxy*-1 : 4-*benzoquinone*, a compound which was also produced by 2 : 6-dimethoxybenzoquinone and by 2 : 3-dimethoxybenzoquinone. The product of hydrolysis in this case was 2 : 5-dihydroxy-3-methoxybenzoquinone, which has been prepared by a different method by Aulin and Erdtman (*Svensk Kem. Tidskr.*, 1937, **49**, 214). The presence of methyl and methoxy-groups in the *para*-position to each other does not, however, invariably or even usually lead to substitution of these groups by the methylamino-group. Thus 3 : 4-dimethoxytoluquinone and 3 : 4 : 6-*trimethoxytoluquinone* reacted to give 3 : 6-*bismethylamino*-4-*methoxy*-2 : 5-*toluquinone*, which on hydrolysis yielded spinulosin, and other examples are recorded later.

All the thirteen methoxy-derivatives of benzoquinone and toluquinone examined gave bismethylamino-derivatives and in all cases the entering methylamino-groups were in the *para*-position to each other. The yields of methylamino-derivatives obtained approximated to those which would be expected from theoretical considerations. Thus in those cases where two methoxy-groups were replaced by two methylamino-groups, *e.g.*, 2 : 5-dimethoxybenzoquinone, 3 : 6-dimethoxy- and 3 : 4 : 6-trimethoxy-toluquinone, the yield approximated to 100%. Where only one methoxy-group was replaced, *e.g.*, methoxy-, 2 : 6- and 2 : 3-dimethoxy-benzoquinone, 3-methoxy- and 3 : 4-dimethoxy-toluquinone, the yield was about 50%. Finally with benzoquinone and toluquinone the yield was of the order of 33%.

The conversion, on treatment with alcoholic methylamine, of 4-methoxytoluquinone (I) into 2 : 5-bismethylamino-1 : 4-benzoquinone (III) with the consequent loss of a methyl group can be explained most readily by supposing that one molecule of methylamine attaches itself to the same carbon atom as the methyl group, with the intermediate formation of (II). Subsequent elimination of the methyl group from (II) as methane, as is known to occur in a number of other reactions (Guareschi, *Gazzetta*, 1918, **48**, II, 83) leads to the formation of (III). A similar reaction would explain the formation of 2 : 5-bismethylamino-3-methoxy-1 : 4-benzoquinone from 4 : 6-dimethoxytoluquinone.



(I.) (II.) (III.)

This suggested mechanism does not explain, however, the yields of methylamino-derivatives obtained in the two cases, *i.e.*, about 30% and 40% respectively instead of the theoretical 100% required by the equation.

Other instances have been reported of the loss of a methyl group from a substituted benzoquinone on treatment with an amine, though not, so far as we are aware, on treatment with monomethylamine. Hoffman (*Ber.*, 1901, **34**, 1558) showed that when an alcoholic solution of dibromothymoquinone is treated with *p*-toluidine, 3 : 6-dibromo-4-*p*-toluidino*iso*propyl-2 : 5-benzoquinone is formed. A similar compound was obtained with aniline in place of toluidine. Boters (*Ber.*, 1902, **35**, 1502), in a continuation of Hoffman's work, found that *m*-toluidine and anisidine react with dibromothymoquinone in an exactly similar fashion in that the methyl group is removed from the nucleus. Similarly *p*-toluidine reacts with dichlorothymoquinone to give 3 : 6-dichloro-4-*p*-toluidino*iso*propyl-2 : 5-benzoquinone with the elimination of the methyl group. On the other hand, monomethylamine behaves towards dibromothymoquinone in a different way, since it yields bismethylaminothymoquinone by the removal of two bromine atoms, but in this case the methyl group remains attached to the nucleus.

The results obtained with nine substituted benzo- and tolu-quinones containing both hydroxyl and methoxyl groups, while interesting in themselves, bear little apparent relationship to each other and do not lend themselves to any obvious generalisations.

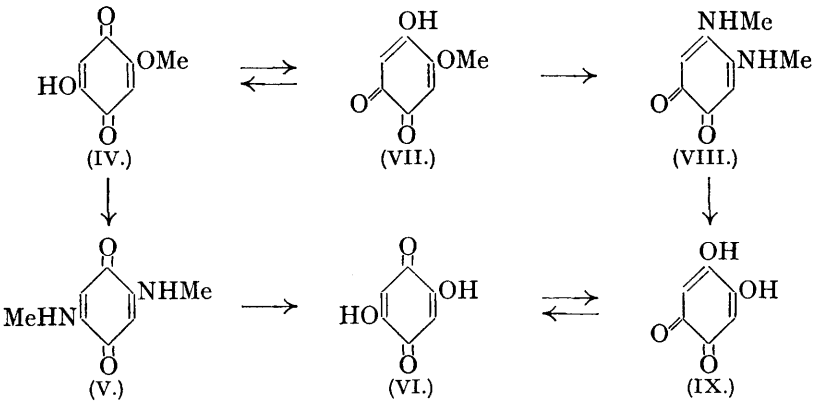
6-Hydroxy-3-methoxytoluquinone, 5-*hydroxy*-2 : 3-*dimethoxybenzoquinone* and 6-*hydroxy*-

3 : 4-*dimethoxytoluquinone* reacted, in each case smoothly, with the replacement by a methylamino-group of the methoxy-group *para* to the hydroxy-group and the formation, respectively, of 3-*methylamino*-6-*hydroxy*-2 : 5-*toluquinone*, 2-*methylamino*-5-*hydroxy*-3-*methoxy*-1 : 4-*benzoquinone* and 3-*methylamino*-6-*hydroxy*-4-*methoxy*-2 : 5-*toluquinone*. The products of hydrolysis of these three methylamino-derivatives were respectively 3 : 6-dihydroxytoluquinone, 2 : 5-dihydroxy-3-methoxybenzoquinone and 3 : 6-dihydroxy-4-methoxytoluquinone (spinulosin).

On the other hand, no methylamino-derivative could be isolated from 4-hydroxy-6-methoxytoluquinone, and 3-hydroxy-4-methoxytoluquinone (*i.e.*, fumigatin, a metabolic product of *Aspergillus fumigatus* Fresenius; Anslow and Raistrick, *Biochem. J.*, 1938, **32**, 687) reacted, with replacement by a methylamino-group of the nuclear hydrogen in position 6, to give 6-*methylamino*-3-*hydroxy*-4-*methoxy*-2 : 5-*toluquinone*, which on hydrolysis gave spinulosin. In all the above cases, where methylamino-derivatives were isolated, they were invariably monomethylamino-derivatives, whereas with the methoxy-compounds of benzo- and tolu-quinone, as described previously, bismethylamino-derivatives invariably resulted.

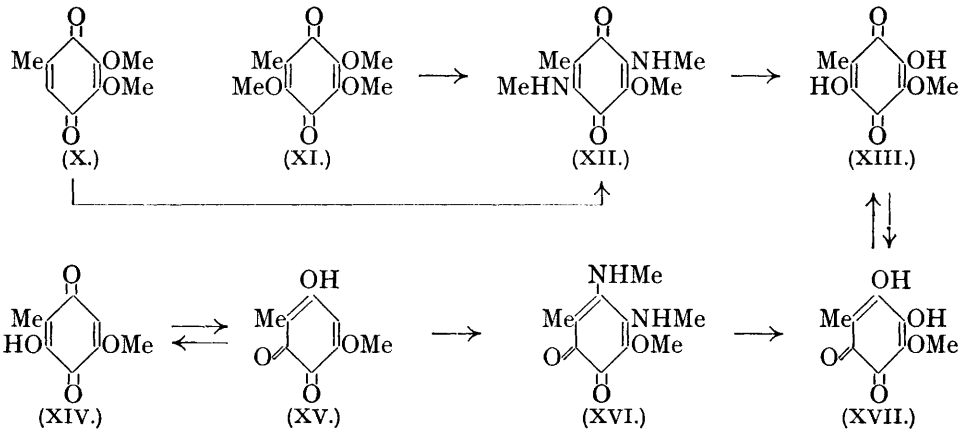
Two dihydroxymonomethoxy-compounds were tested, *viz.*, 2 : 5-dihydroxy-3-methoxybenzoquinone and 3 : 6-dihydroxy-4-methoxytoluquinone. With these substances no replacement of any groups by methylamine was evident, since bis-methylamine salts resulted in each case.

The two remaining quinones, 2-hydroxy-5-methoxy-1 : 4-benzoquinone (IV) and 6-hydroxy-4-methoxy-2 : 5-toluquinone (XIV), reacted with methylamine in a somewhat unusual manner. The former gave a mixture of two different bismethylamino-benzoquinones, each having the empirical formula $C_8H_{10}O_2N_2$ and each giving 2 : 5-dihydroxy-1 : 4-benzoquinone (VI) on alkaline hydrolysis. One of these methylamino-derivatives melts at 285—286°, is insoluble in water and only very slightly soluble in 2N-sulphuric acid, and is the same product as was obtained in theoretical yield by treating 2 : 5-dimethoxy-1 : 4-benzoquinone with methylamine (see p. 1447). This methylamino-derivative is doubtless 2 : 5-bismethylamino-1 : 4-benzoquinone (V). Its slight solubility in 2N-sulphuric acid could then be readily explained, since each C=O group might be expected to neutralise its adjacent methylamino-group. The second methylamino-derivative does not melt below 360°, but is readily soluble in dilute acid and even in water. Its formation and properties are readily explained if it be assumed that 2-hydroxy-5-methoxy-1 : 4-benzoquinone (IV), on treatment with alcoholic methylamine, reacts in its tautomeric form (VII) to give 4 : 5-*bismethylamino*-1 : 2-*benzoquinone* (VIII), which on hydrolysis gives (IX). The fact that both (V) and (VIII) give the same product on hydrolysis then follows, since (VI) and (IX) are tautomerides.



Finally, 6-hydroxy-4-methoxy-2 : 5-toluquinone (XIV) gave a bismethylamino-derivative which, as we have previously reported (Anslow and Raistrick, *Biochem. J.*, 1938, **32**, 803), gives on acid hydrolysis a good yield of spinulosin and affords a very convenient method

for the synthesis of this substance. This methylamino-derivative, $C_{10}H_{14}O_3N_2$, might be expected to be identical with the methylamino-derivative, of the same empirical formula and also giving spinulosin on acid hydrolysis, which was obtained by treating either 3 : 4-dimethoxy-2 : 5-toluquinone (X) or 3 : 4 : 6-trimethoxy-2 : 5-toluquinone (XI) with alcoholic methylamine. In fact the two methylamino-derivatives are quite distinct chemical substances: the derivative from 6-hydroxy-4-methoxytoluquinone gives a pure blue solution in chloroform, is readily soluble in cold N/10-sulphuric acid, and melts at 228°, that from 3 : 4-dimethoxytoluquinone and 3 : 4 : 6-trimethoxytoluquinone gives a purple solution in chloroform, is insoluble even in 2N-sulphuric acid, and melts at 231°, and a mixture of the two substances melts at 219°. These facts can be readily explained on the same lines as are given above for the formation of two different bismethylamino-derivatives from 2-hydroxy-5-methoxybenzoquinone. Thus, since it is difficult, if not indeed impossible, to conceive of 3 : 4 : 6-trimethoxytoluquinone (XI) undergoing any tautomeric change, the substance obtained when this quinone or 3 : 4-dimethoxytoluquinone (X) reacts with methylamine must be 3 : 6-bismethylamino-4-methoxy-2 : 5-toluquinone (XII) and the insolubility of this substance in 2N-sulphuric acid is then to be expected, since each C=O group is adjacent to a methylamino-group. 6-Hydroxy-4-methoxytoluquinone (XIV) must then react with methylamine in its tautomeric form (XV) to give 5 : 6-bismethylamino-4-methoxy-2 : 3-toluquinone (XVI). The fact that both (XII) and (XVI) give spinulosin on acid hydrolysis follows, since (XII) would give (XIII) and (XVI) would give (XVII) on hydrolysis and (XIII) and (XVII) are tautomerides.



From the results described above it will be seen that four derivatives of 4-methoxytoluquinone, *viz.*, 3-hydroxy-4-methoxytoluquinone, 6-hydroxy-4-methoxytoluquinone, 3 : 6-dihydroxy-4-methoxytoluquinone and 6-hydroxy-3 : 4-dimethoxytoluquinone, are included in the hydroxymethoxyquinones examined. In none of these cases was there any indication of the substitution of methylamine for the methyl group and the methoxyl group in the *para*-position to it such as is described previously for 4-methoxytoluquinone and 4 : 6-dimethoxytoluquinone.

Further experiments are in progress on the action of methylamine on a number of mono- and di-hydroxy-derivatives of benzo- and tolu-quinone, the results of which will be reported at a later date.

Experimental.

The method adopted throughout the following experiments was to dissolve the quinone under examination in a suitable volume of cold or hot ethanol and to add to the solution a large excess of a 33% w./w. solution of monomethylamine in ethanol. The methylamino-derivative which separated was purified and identified.

(I) Benzoquinone (5 g.) was dissolved in ethanol (250 ml.), and alcoholic methylamine (25 ml.) added at room temperature. The mixture became intensely purple-brown and was kept

at 0° for 3 days. The amorphous, dull purple product separating (1·6 g.) was removed by filtration, washed with ethanol, dried, sublimed in a high vacuum at 160°, and crystallised from ethanol, forming long silky cerise needles of 2 : 5-*bismethylamino*-1 : 4-*benzoquinone*, m. p. 284—286° (decomp.) (Found: C, 58·0; H, 6·0; N, 16·8. $C_8H_{10}O_2N_2$ requires C, 57·8; H, 6·1; N, 16·9%). It dissolves in chloroform to give a dull orange solution, is slightly soluble in cold 2N- and readily soluble in 5N-sulphuric acid, giving a reddish-purple solution, and almost insoluble in cold N-sodium hydroxide.

(II) Methoxy-1 : 4-benzoquinone (Mulhauser, *Annalen*, 1881, **207**, 251) (0·1 g., m. p. 145—146°) was dissolved in boiling ethanol (2 ml.), and alcoholic methylamine (0·5 ml.) added. The solution immediately became intensely purple-brown and after a few seconds purple needles began to separate. After 1½ hours these were filtered off, washed, dried (0·05 g.), sublimed in a high vacuum at 160°, and then crystallised from ethanol, forming cerise needles, m. p. 285—287°, not depressed on admixture with 2 : 5-bismethylamino-1 : 4-benzoquinone prepared from benzoquinone.

(III) 2 : 5-Dimethoxy-1 : 4-benzoquinone (Knoevenagel and Bückel, *Ber.*, 1901, **34**, 3996) (0·3 g., m. p. 303°) was suspended in boiling ethanol (12 ml.), and alcoholic methylamine (1·5 ml.) added. The mixture shortly became reddish-purple and was shaken at frequent intervals. The quinone slowly dissolved and reddish-purple silky needles separated. These were filtered off overnight, dried (0·25 g.), and crystallised first from ethanol and then from chloroform, forming silky cerise needles, m. p. 284—287°, not depressed on admixture with 2 : 5-bismethylamino-1 : 4-benzoquinone prepared from benzoquinone.

(IV) 2 : 6-Dimethoxy-1 : 4-benzoquinone (1·0 g., m. p. 254—255°), prepared by the oxidation of pyrogallol trimethyl ether (Ullmann, *Annalen*, 1903, **327**, 116) with alcoholic nitric acid (Graebe and Hess, *Annalen*, 1905, **340**, 237), was suspended in boiling ethanol (50 ml.), and alcoholic methylamine (5 ml.) added. The mixture, which became red, was shaken frequently; the quinone slowly dissolved. After standing for 24 hours, the steel-grey needles which had separated were filtered off, washed with ethanol, and dried (0·51 g.). The substance, 2 : 5-*bismethylamino*-3-*methoxy*-1 : 4-*benzoquinone*, was sublimed in a high vacuum at 140°, and crystallised from ethanol, forming steel-grey needles, m. p. 234° (Found: C, 55·1; H, 6·3; N, 13·9. $C_9H_{12}O_3N_2$ requires C, 55·1; H, 6·2; N, 14·3%). It gives a deep purple (permanganate) solution in chloroform and is insoluble in 2N- and sparingly soluble in cold 5N-sulphuric acid to give a violet-blue solution. It is almost insoluble in cold N-sodium hydroxide. Its constitution follows from the fact that on acid hydrolysis it gives 2 : 5-dihydroxy-3-methoxy-1 : 4-benzoquinone (see section XII), identified by its m. p. 159—160°, alone or in admixture with an authentic specimen, by its colour reactions and by analysis (Found: C, 49·1; H, 3·8. Calc. for $C_7H_6O_5$: C, 49·4; H, 3·6%).

(V) 2 : 3-Dimethoxy-1 : 4-benzoquinone (Baker and Smith, J., 1931, 2547) (0·1 g., m. p. 66—67°), prepared by decarboxylation of 2 : 5-dihydroxy-3 : 4-dimethoxybenzoic acid and oxidation of the resulting quinol (Baker and Savage, J., 1938, 1604), was dissolved in ethanol (2 ml.) and to the cold solution 0·5 ml. of methylamine was added. The solution, which immediately became deep purple-brown, deposited purple-black crystals after 15 minutes, which were separated after 24 hours, washed and dried. This crude 2 : 5-bismethylamino-3-methoxy-1 : 4-benzoquinone (0·04 g.), m. p. 231—232°, was sublimed in a high vacuum at 140°, giving 0·025 g. of a crystalline sublimate, m. p. 234°, not depressed on admixture with 2 : 5-bismethylamino-3-methoxybenzoquinone prepared from 2 : 6-dimethoxybenzoquinone (see previous section). The substance, which crystallises from ethanol in steel-grey crystals, also shows the same solubilities and colours in chloroform, 2N- and 5N-sulphuric acid, and cold sodium hydroxide as the product from 2 : 6-dimethoxybenzoquinone.

(VI) It was shown by Fichter (*Annalen*, 1908, **361**, 400) that toluquinone, on treatment with methylamine, gives a bismethylamino-derivative; this must be 3 : 6-bismethylamino-2 : 5-toluquinone, since it gives on hydrolysis a dihydroxytoluquinone which on methylation yields 3 : 6-dimethoxytoluquinone (Anslow, Ashley, and Raistrick, J., 1938, 439). The yield of crude methylamino-compound is small (12·2 g. of toluquinone, dissolved in 100 ml. of ethanol and treated with 20 ml. of alcoholic methylamine, gave 2·6 g. of crude product) and the substance is very impure. It was purified by sublimation in a high vacuum at 140—150°, followed by crystallisation from ethanol; the purple-brown crystals had m. p. 231°. Fichter (*loc. cit.*) gives 235°. It is readily soluble in chloroform, giving a crimson solution. It is insoluble in N-sodium hydroxide, slightly soluble in 2N- and readily soluble in cold 5N-sulphuric acid, giving a deep purple (permanganate) solution.

(VII) 3-Methoxytoluquinone (Henrich and Nachtigall, *Ber.*, 1903, **36**, 899) (0·15 g., m. p.

149—150°) was dissolved in boiling ethanol (3 ml.), and alcoholic methylamine (0·75 ml.) added. The solution immediately became intensely reddish-purple and crystals quickly separated. After 3 hours these were filtered off, washed, and dried (0·08 g., m. p. 230—231°). A portion was sublimed in a high vacuum at 140—150°. The purple-brown sublimate melted at 230—231° alone or in admixture with 3 : 6-bismethylamino-2 : 5-toluquinone prepared from toluquinone. It also behaved in the same way towards chloroform, N-sodium hydroxide and 5N-sulphuric acid.

(VIII) 4-Methoxytoluquinone (5 g., m. p. 172—173°) was dissolved in boiling ethanol (250 ml.), and alcoholic methylamine (25 ml.) added. The solution immediately became intensely brownish-purple. After 24 hours the purple needles which had separated were filtered off, washed with ethanol, and dried (1·6 g., m. p. 269—270°). The crude material was crystallised thrice from ethanol, once from chloroform and once from toluene, giving long cerise needles of 2 : 5-bismethylamino-1 : 4-benzoquinone, m. p. 285—286° (decomp.), not depressed on admixture with a specimen prepared from benzoquinone (see section I) (Found : C, 57·8; H, 6·2; N, 16·7%; OMe, nil). The same product was obtained in the same yield from 4-methoxytoluquinone prepared either from toluquinone (Ashley, J., 1937, 1471) or by the method of Luff, Perkin, and Robinson (J., 1910, **97**, 1137) and hence the possibility is excluded that the methylamino-derivative arises from benzoquinone present as an impurity in the starting material.

The constitution of the methylamino-derivative was fully established as follows : 0·8 g. was boiled for 4 minutes with 2N-sodium hydroxide (80 ml.), the solution cooled, acidified, and extracted with ether, and the extract dried and evaporated to dryness. The residue was sublimed in a high vacuum and the crystalline sublimate (0·25 g.) was recrystallised from toluene, giving 0·22 g. of dark orange needles (2 : 5-dihydroxy-1 : 4-benzoquinone), which began to darken at 170° and became progressively darker and finally black without melting at 300° (Found : C, 51·4; H, 3·0; OMe, nil. Calc. for $C_6H_4O_4$: C, 51·4; H, 2·9%). The quinone gives a deep cherry-red colour with concentrated sulphuric acid, a cerise colour with 2N-sodium hydroxide, and a dark reddish-brown colour with ferric chloride in alcohol. The quinone (0·15 g.) was shaken with a freshly prepared solution of sodium hyposulphite (3 g.) in water (15 ml.). The almost colourless solution quickly obtained was extracted with ether, the extract dried and evaporated to dryness, and the residue (0·13 g.) sublimed in a high vacuum. The colourless crystalline sublimate (1 : 2 : 4 : 5-tetrahydroxybenzene) melted at 232—233° with some decomposition from 200° (Nietzki and Schmidt, *Ber.*, 1888, **21**, 2377, give 215—220°) (Found : C, 50·5; H, 4·3. Calc. for $C_6H_6O_4$: C, 50·7; H, 4·3%). 1 : 2 : 4 : 5-Tetrahydroxybenzene gives with concentrated sulphuric acid a yellow colour quickly changing to emerald-green. With 2N-sodium hydroxide it gives a yellow solution quickly changing to olive-green, then brown and finally cerise, and a dark reddish-brown colour with ferric chloride in alcohol. 1 : 2 : 4 : 5-Tetrahydroxybenzene was heated with acetic anhydride and a little concentrated sulphuric acid to give 1 : 2 : 4 : 5-tetra-acetoxybenzene, which formed colourless prisms from ethanol, m. p. 226—227°, not depressed on admixture with a specimen prepared from 2 : 5-bisdimethylaminobenzoquinone (Mylius, *Ber.*, 1885, **18**, 463; Kehrmann, *Ber.*, 1890, **23**, 897). Jackson and Beggs (*J. Amer. Chem. Soc.*, 1914, **36**, 1216) give m. p. 226—227° (Found : C, 54·2; H, 4·6. Calc. for $C_{14}H_{14}O_8$: C, 54·2; H, 4·6%).

(IX) 6-Methoxytoluquinone (Majima and Okazaki, *Ber.*, 1916, **49**, 1490; Anslow, Ashley, and Raistrick, J., 1938, 439) (1·0 g., m. p. 19—20°) was dissolved in cold ethanol (10 ml.), and alcoholic methylamine (5 ml.) added. The solution, which became intensely purple-brown, quickly began to deposit crystals. After standing overnight, these were separated by filtration, washed, dried (0·2 g.), sublimed in a high vacuum at 140—150°, and crystallised from ethanol. The purple-brown crystals had m. p. 230°, alone or in admixture with 3 : 6-bismethylamino-2 : 5-toluquinone prepared from toluquinone (Found : C, 60·4; H, 6·6; N, 15·4; OMe, nil. Calc. for $C_9H_{12}O_2N_2$: C, 60·0; H, 6·7; N, 15·6%).

(X) 3 : 6-Dimethoxytoluquinone (Anslow, Ashley, and Raistrick, J., 1938, 439) (0·05 g., m. p. 112°) was dissolved in warm ethanol (2 ml.). To the golden-orange solution was added alcoholic methylamine (0·25 ml.); the mixture became deep crimson and almost immediately purple-brown irregular leaflets separated (0·04 g., m. p. 230°). After sublimation in a high vacuum at 140—150° the purple-brown sublimate melted at 230—231°, alone or in admixture with 3 : 6-bismethylamino-2 : 5-toluquinone prepared from toluquinone. It also behaved in the same way towards chloroform, N-sodium hydroxide and 5N-sulphuric acid.

(XI) 3 : 4-Dimethoxytoluquinone (Anslow, Ashley, and Raistrick, *loc. cit.*) (2·36 g.) was dissolved in ethanol (24 ml.), and alcoholic methylamine (12 ml.) added at room temperature. The mixture immediately became dark brown and crystals quickly formed. They were

separated after 3 hours (0·92 g., m. p. 227—228°) and purified for analysis by sublimation in a high vacuum at 130° and crystallisation from ethanol. 3 : 6-*Bismethylamino-4-methoxy-2* : 5-*toluquinone* crystallised in glistening purple-grey plates, m. p. 231° with partial sublimation but without obvious decomposition (Found : C, 57·2; H, 6·5; N, 13·0. $C_{10}H_{14}O_3N_2$ requires C, 57·1; H, 6·7; N, 13·3%). The substance gave a deep purple solution in chloroform but was insoluble in 2N-sodium hydroxide, 2N- and 5N-sulphuric acid. 0·5 G. was hydrolysed by boiling for 3 minutes with 2N-sulphuric acid (50 ml.); an intensely purple solution was formed and quickly changed to brownish-red. On cooling, spinulosin (3 : 6-dihydroxy-4-methoxy-2 : 5-toluquinone) (0·36 g.) crystallised; it was identified by its m. p. 201°, alone or in admixture with an authentic specimen, by its diacetate, canary-yellow rods from ethanol, m. p. 139·5° alone or mixed with an authentic specimen (Birkinshaw and Raistrick, *Phil. Trans.*, 1931, *B*, **220**, 250), and by analysis (Found : C, 52·1; H, 4·3. Calc. for $C_8H_8O_5$: C, 52·2; H, 4·4%).

(XII) 4 : 6-Dimethoxytoluquinone (Anslow, Ashley, and Raistrick, J., 1938, 439; Aulin and Erdtman, *Svensk Kem. Tidsk.*, 1938, **50**, 42) (1 g., m. p. 125°) was dissolved in ethanol (20 ml.) at 60°, and alcoholic methylamine (5 ml.) added. The solution, which quickly became deep blood-red, deposited, on standing overnight, purple-black crystals (0·4 g., m. p. 228°), which were sublimed at 140° in a high vacuum. The sublimate crystallised from ethanol in glistening steel-grey needles of 2 : 5-bismethylamino-3-methoxy-1 : 4-benzoquinone, m. p. 234° after shrinking from 230°, not depressed on admixture with a specimen prepared from 2 : 6-dimethoxybenzoquinone (see section IV) (Found : C, 55·2; H, 5·9; N, 13·9%).

The constitution of the methylamino-derivative was established as follows : 0·35 g. was boiled for 1½ minutes with 5N-sulphuric acid (14 ml.). The hot solution, initially purple and quickly becoming reddish-brown, was cooled, diluted with an equal volume of water, and extracted with ether. The extract was dried and evaporated to dryness, and the residue sublimed in a high vacuum at 100°. The sublimate (0·17 g.) crystallised from toluene in coppery leaflets of 2 : 5-dihydroxy-3-methoxy-1 : 4-benzoquinone, which were resublimed, m. p. 159—160°, alone or in admixture with an authentic specimen kindly supplied by Dr. H. Erdtman. This specimen was prepared by Aulin and Erdtman (*Svensk Kem. Tidskr.*, 1937, **49**, 214) by a method which leaves no doubt as to its orientation (Found : C, 49·4; H, 3·6; OMe, 18·2. Calc. for $C_7H_6O_5$: C, 49·4; H, 3·6; OMe, 18·3%). It gives colour reactions which are almost identical with those given by its toluene homologue, spinulosin—pure blue with cold concentrated sulphuric acid, purple with 2N-sodium hydroxide, and deep rich brown with ferric chloride in alcohol.

2 : 5-*Diacetoxy-3-methoxy-1* : 4-*benzoquinone* was prepared by heating the quinone (0·08 g.) for a short time with acetic anhydride (1 ml.) and one drop of concentrated sulphuric acid. The cooled mixture was poured into ice-water and the solid separating was collected, dried, and sublimed in a high vacuum at 65°. The lemon-yellow prisms (0·05 g.) had m. p. 77°, alone or in admixture with a specimen of the diacetate prepared from 2 : 5-dihydroxy-3-methoxybenzoquinone supplied by Dr. Erdtman (Found : C, 52·0; H, 4·0; OMe, 12·2. $C_{11}H_{10}O_7$ requires C, 52·0; H, 4·0; OMe, 12·2%).

(XIII) 3 : 4 : 6-Trimethoxytoluquinone (for method of preparation, see below) (0·2 g.) was dissolved in warm ethanol (2 ml.), and alcoholic methylamine (1 ml.) added. The intensely purple-brown solution quickly deposited purple-black crystals (0·19 g.) of 3 : 6-bismethylamino-4-methoxy-2 : 5-toluquinone, which were sublimed in a high vacuum at 130° and crystallised from ethanol. The dark purple-grey plates had m. p. 230°, alone or in admixture with 3 : 6-bismethylamino-4-methoxytoluquinone prepared from 3 : 4-dimethoxytoluquinone (see section XI) (Found : C, 57·2; H, 6·7; N, 13·1%).

3 : 4 : 6-*Trimethoxy-2* : 5-*toluquinone* (*Dimethyl Ether of Spinulosin*).—3 : 6-Dihydroxy-4-methoxytoluquinone (spinulosin; Birkinshaw and Raistrick, *Phil. Trans.*, 1931, *B*, **220**, 245; Anslow and Raistrick, *Biochem. J.*, 1938, **32**, 803) (0·6 g.) was suspended in ether, and ethereal diazomethane added in small amounts until the initial vigorous reaction ceased. The solution was evaporated to dryness, the dark red residue (0·65 g.) redissolved in ether, and the solution extracted with successive small amounts of p_H 8·0 buffer solution until no further colour was extracted. The extracted ethereal solution on evaporation gave 0·51 g. of deep red needles of 3 : 4 : 6-*trimethoxytoluquinone*, which were sublimed in a high vacuum at 60—70° and crystallised from ethanol, forming reddish-orange needles, m. p. 80°, which gave a pure deep blue colour with cold concentrated sulphuric acid but no immediate colour with 2N-sodium hydroxide, although a purple colour slowly developed on standing. The quinone also gave no marked colour with ferric chloride in alcoholic or aqueous solution (Found : C, 56·6; H, 5·6; OMe, 43·7. $C_{10}H_{12}O_5$

requires C, 56·6; H, 5·7; 3OMe, 43·9%). The same trimethyl ether was formed by methylating spinulosin with methyl sulphate and potassium carbonate in acetone solution, but the yield was small.

2 : 5-*Dihydroxy*-3 : 4 : 6-*trimethoxytoluene*. 3 : 4 : 6-Trimethoxytoluquinone (0·15 g.), shaken with a solution of sodium hyposulphite (4 g.) in water (20 ml.), slowly dissolved to give a colourless solution, which was extracted with ether. The dried extract on evaporation gave a crystalline residue, which was sublimed in a high vacuum at 75°, yielding colourless irregular leaflets of 2 : 5-*dihydroxy*-3 : 4 : 6-*trimethoxytoluene*. Yield, almost theoretical, m. p. 82—83°. The quinol gives with cold concentrated sulphuric acid an immediate bright yellow colour, changing after ½ hour to a dark olive-green, with 2N-sodium hydroxide an immediate yellowish-green colour, fading to colourless in a few seconds, and with ferric chloride in aqueous or alcoholic solution a golden-yellow colour (Found: C, 56·1; H, 6·6; OMe, 43·5. $C_{10}H_{14}O_5$ requires C, 56·1; H, 6·6; 3OMe, 43·5%).

(XIV) 2-Hydroxy-5-methoxy-1 : 4-benzoquinone (for method of preparation, see below) (0·5 g.) was dissolved in boiling ethanol (15 ml.), and methylamine (2·5 ml.) added. The mixture, now reddish-purple, was kept at room temperature for 3 hours with occasional shaking. The dark purple rods formed (0·32 g.) were separated by filtration and the mother-liquor after 2 days deposited 0·12 g. of purple-red needles, which, after sublimation in a high vacuum and crystallisation from ethanol, formed long cerise needles, m. p. 285—286°, of 2 : 5-bismethylamino-1 : 4-benzoquinone (see section I). The dark purple rods (0·32 g.) were recrystallised from ethanol and the substance, probably 4 : 5-*bismethylamino*-1 : 2-*benzoquinone*, was thus obtained as large, dark purple rods which did not melt below 360° (Found: C, 58·1; H, 6·1; N, 16·9; OMe, nil. $C_8H_{10}O_2N_2$ requires C, 57·8; H, 6·1; N, 16·9%). The substance was readily soluble in water, giving a carmine-coloured solution, but was only slightly soluble in chloroform. On hydrolysis with boiling 2N-sodium hydroxide it gave 2 : 5-dihydroxy-1 : 4-benzoquinone, which was identified by its colour reactions and by reduction to 1 : 2 : 4 : 5-tetrahydroxybenzene; this, on acetylation, gave 1 : 2 : 4 : 5-tetra-acetoxybenzene, m. p. 226—227°, not depressed on admixture with an authentic specimen (see section VIII).

2-*Hydroxy*-5-*methoxy*-1 : 4-*benzoquinone*. 1 : 2 : 4-Triacetoxy-5-methoxybenzene (5 g.), prepared by a Thiele–Winter acetylation of methoxybenzoquinone (Erdtman, *Proc. Roy. Soc.*, 1933, *A*, **143**, 186), was boiled for ¾ hour in an atmosphere of nitrogen with 37·5 ml. of a mixture of methanol (60 ml.) and concentrated sulphuric acid (2 ml.). Water was added, and the methanol removed by distillation in a vacuum. The quinol (2·56 g.) was extracted with ether. The crude quinol (1·1 g.) was dissolved in 110 ml. of p_H 8·0 buffer solution (M-potassium dihydrogen phosphate, 50 ml.; N-sodium hydroxide, 46·8 ml.; water to 100 ml.) and aerated vigorously for 10 minutes. The intensely blood-red solution was acidified with concentrated hydrochloric acid (16·5 ml.). 2-*Hydroxy*-5-*methoxybenzoquinone* quickly crystallised (0·60 g.); a further 0·45 g. of slightly impure quinone was obtained by ether extraction of the filtrate. It formed large, orange-brown, rectangular leaflets from ethanol, m. p. 179° (decomp.) after darkening and softening from 171° (Found: C, 54·5; H, 4·0; OMe, 20·05. $C_7H_6O_4$ requires C, 54·5; H, 3·9; OMe, 20·1%). The quinone sublimes readily in a high vacuum at 100°. It gives a dull red colour with cold concentrated sulphuric acid, a dull red colour, changing to cerise, with 2N-sodium hydroxide, and a deep reddish-brown colour in ethanol with aqueous ferric chloride.

1 : 2 : 4-*Trihydroxy*-5-*methoxybenzene*. The above quinone (0·15 g.), shaken with a solution of sodium hyposulphite (1·5 g.) in water (7·5 ml.), quickly dissolved to give an almost colourless solution, which was extracted with ether. On removal of the solvent a colourless oil, which rapidly crystallised, was obtained; it sublimed in a high vacuum at 100° in colourless feathery needles (yield, almost theoretical), m. p. 133° (Found: C, 53·9; H, 5·2; OMe, 19·6. $C_7H_8O_4$ requires C, 53·8; H, 5·2; OMe, 19·9%). This *quinol* gives with cold concentrated sulphuric acid a yellow colour, quickly changing to apple-green and finally to dark olive-green, with 2N-sodium hydroxide a green colour, changing through blue to brown and finally to dull red, and with aqueous ferric chloride in ethanol a deep reddish-brown colour.

2-*Acetoxy*-5-*methoxybenzoquinone*. 2-Hydroxy-5-methoxybenzoquinone does not appear to undergo the usual Thiele–Winter reaction. The quinone (0·2 g.) was added to 2 ml. of a mixture of acetic anhydride (20 ml.) and concentrated sulphuric acid (1 ml.). The orange solution changed to yellow in a few minutes and was poured into ice-water after 3 hours. The lemon-yellow needles separating (0·04 g.) were sublimed in a high vacuum at 90—100°; a further small quantity was obtained by ether extraction of the acetylation filtrate. The lemon-yellow rectangular plates had m. p. 124° (Found: C, 55·2; H, 4·1; OMe, 15·8. $C_9H_8O_5$ requires C, 55·1; H, 4·1; OMe, 15·8%).

(XV) 5-Hydroxy-2 : 3-dimethoxybenzoquinone (for method of preparation, see below) (0·4 g.) was dissolved in cold ethanol (4 ml.), and methylamine (2 ml.) added. The colour immediately changed from orange-red to purple-red and dark purple, feathery needles quickly formed. These were separated after 3 hours, washed with ethanol, and dried (0·39 g.); m. p. 228—230° (decomp.) (Found: C, 50·5; H, 6·6; N, 13·1; OMe, 15·2. $C_8H_9O_4N,NH_2Me$ requires C, 50·4; H, 6·6; N, 13·1; OMe, 14·5%). The substance, which is the *monomethylamine* salt of *2-methylamino-5-hydroxy-3-methoxy-1 : 4-benzoquinone*, is insoluble in chloroform, but is readily soluble in water. On hydrolysis with boiling 5N-sulphuric acid it gave in good yield 2 : 5-dihydroxy-3-methoxybenzoquinone, which was identified by its colour reactions and m. p. 160—161°, not depressed on admixture with an authentic specimen (see section XII).

The methylamine salt (0·107 g.; 1/2000 g.-mol.) was dissolved in water (5 ml.) and to the clear, intensely purple (permanganate) solution N/10-hydrochloric acid (5 ml.) was added. *2-Methylamino-5-hydroxy-3-methoxybenzoquinone* (0·08 g.) immediately separated in purple-black hexagonal plates, m. p. 179° (decomp.), which were readily soluble in chloroform and N/10-sodium hydroxide, giving a purple solution in each case, and gave an olive-brown colour with ferric chloride in ethanol (Found: N, 7·65. $C_8H_9O_4N$ requires N, 7·65%).

5-Hydroxy-2 : 3-dimethoxy-1 : 4-benzoquinone. 2 : 3-Dimethoxyquinol (4·9 g.), prepared by the decarboxylation of 2 : 5-dihydroxy-3 : 4-dimethoxybenzoic acid (Baker and Savage, J., 1938, 1604), was dissolved in water (225 ml.), and the theoretical volume of M/6-ferric chloride quickly added. The resulting orange-red solution was extracted twice with an equal volume of chloroform. On removal of the solvent from the dried solution 2 : 3-dimethoxybenzoquinone (3·85 g.) remained as a reddish oil which quickly crystallised. This was dissolved in 20 ml. of a mixture of acetic anhydride (100 ml.) and concentrated sulphuric acid (5 ml.) and after 3 days was poured into ice-water. The oil which separated soon crystallised. The light brown crystals were dried and washed with anhydrous ether (25 ml.), giving 4·5 g. of 1 : 4 : 5-triacetoxy-2 : 3-dimethoxybenzene in colourless crystals, m. p. 95—97° (Erdtman, *Proc. Roy. Soc.*, 1933, *A*, 143, 187, gives 96—97°). The triacetate (1 g.) was boiled for ¾ hour in an atmosphere of nitrogen with 10 ml. of a mixture of methanol (60 ml.) and concentrated sulphuric acid (2 ml.). After removal of the solvent the crude quinol obtained by ether extraction of the hydrolysis solution was dissolved in 50 ml. of p_H 8·0 buffer solution and vigorously aerated for ½ hour. The resulting intensely purple-red solution was acidified with concentrated hydrochloric acid (7·5 ml.) and extracted with ether. On removal of the solvent *5-hydroxy-2 : 3-dimethoxybenzoquinone* (0·42 g.) remained as an orange solid, which was sublimed in a high vacuum at 85° and crystallised from light petroleum (b. p. 60—80°), forming long reddish-orange rods, m. p. 125—126° after softening from 115° (Found: C, 52·1; H, 4·3; OMe, 33·5. $C_8H_8O_5$ requires C, 52·2; H, 4·4; 2OMe, 33·7%). The quinone gives an intense pure blue colour with cold concentrated sulphuric acid, a magenta colour, changing after 5 minutes to a stable purple, with 2N-sodium hydroxide, and an intense reddish-brown colour with aqueous ferric chloride in alcoholic solution.

1 : 4 : 5-Trihydroxy-2 : 3-dimethoxybenzene. 5-Hydroxy-2 : 3-dimethoxybenzoquinone (0·1 g.) was reduced with sodium hyposulphite (1 g. in 5 ml. of water). After extraction with ether and removal of the solvent the crystalline residue was sublimed in a high vacuum at 110°, giving *1 : 4 : 5-trihydroxy-2 : 3-dimethoxybenzene* (0·09 g.) in colourless needles, m. p. 157—158° (Found: C, 51·7; H, 5·4; OMe, 33·0. $C_8H_{10}O_5$ requires C, 51·6; H, 5·4; 2OMe, 33·35%). The substance gives with cold concentrated sulphuric acid a canary-yellow colour, quickly changing through yellowish-green to emerald-green, with 2N-sodium hydroxide an emerald-green, changing through greenish-brown to cerise, and with aqueous ferric chloride in ethanol an intense reddish-brown (iodine) colour.

(XVI) 2 : 5-Dihydroxy-3-methoxy-1 : 4-benzoquinone (Aulin and Erdtman, *Svensk Kem. Tidskr.*, 1937, 49, 214; Anslow and Raistrick, section XII) (0·3 g.) was dissolved in ethanol (5 ml.) and to the cool solution methylamine (1·5 ml.) was added. There was an immediate precipitation of a violet micro-crystalline product, which was separated, washed, and dried (0·36 g.); m. p. 214° (decomp.) (Found: C, 45·6; H, 7·1; N, 11·9. $C_7H_6O_5,2NH_2Me$ requires C, 46·5; H, 7·0; N, 12·1%). The product is readily soluble in water, giving a violet solution, but is insoluble in chloroform. That the product is the *bismethylamine* salt of *2 : 5-dihydroxy-3-methoxybenzoquinone* is shown by the fact that it dissolves in cold N-sulphuric acid to give a reddish-brown solution, from which ether extracts an almost quantitative yield of 2 : 5-dihydroxy-3-methoxybenzoquinone.

(XVII) 3-Hydroxy-4-methoxy-2 : 5-toluquinone, *i.e.*, fumigatin, a metabolic product of *Aspergillus fumigatus* Fresenius (Anslow and Raistrick, *Biochem. J.*, 1938, 32, 687) (0·3 g.) was dissolved in cold ethanol (3 ml.), and alcoholic methylamine (1·5 ml.) added. The solution

immediately became intensely purple and after 4 hours the purple-brown needles (0·13 g.) separating were filtered off, dried, and sublimed in a high vacuum at 120°. The sublimate (0·09 g.) crystallised from ethanol in coppery-purple leaflets of 6-*methylamino*-3-*hydroxy*-4-*methoxy*-2 : 5-*toluquinone*, m. p. 213—214° (Found : C, 54·7; H, 5·7; N, 6·7. $C_9H_{11}O_4N$ requires C, 54·8; H, 5·6; N, 7·1%). The substance gives a deep purple solution in chloroform. It is not decomposed or dissolved by cold 2N-sulphuric acid and is almost insoluble in cold 5N-sulphuric acid. It is readily soluble in cold N/10-sodium hydroxide, giving a deep violet solution. An alcoholic solution gives with aqueous ferric chloride an intense olive-green colour (cf. 3-methylamino-6-hydroxy-4-methoxy-2 : 5-toluquinone, section XXII). It is readily hydrolysed by boiling for 1 minute with 2N-sulphuric acid to give in good yield spinulosin (3 : 6-dihydroxy-4-methoxy-2 : 5-toluquinone), which was identified by its m. p., 201°, not depressed on admixture with an authentic specimen, and by its colour reactions.

(XVIII) 6-Hydroxy-4-methoxy-2 : 5-toluquinone (Konya, *Monatsh.*, 1900, **21**, 422; Pollak and Solomonica, *ibid.*, 1901, **22**, 1008; Anslow, Ashley, and Raistrick, J., 1938, 439) (1·25 g., m. p. 203—204°) was dissolved in boiling ethanol (60 ml.), the solution cooled to 35° without shaking, and alcoholic methylamine (6 ml.) added. The solution became intensely purple and the purple-black crystals separating were filtered off overnight, washed, and dried (0·6 g., m. p. 214—215°). The substance could be crystallised from ethanol or toluene but was most conveniently purified as follows : 0·1 g. was dissolved in cold N-sulphuric acid (30 ml.), in which it was readily soluble, giving an intense blue-violet solution. This was filtered and N-sodium hydroxide (30·3 ml.) was added to the filtrate. Long olive-green rods separated, m. p. 228°. The m. p. of a mixture with the bismethylamino-derivative from 3 : 4-dimethoxytoluquinone, m. p. 231° (see section XI), was 219° (Found for air-dried material : C, 52·7; H, 7·3; N, 12·5. $C_{10}H_{14}O_3N_2,H_2O$ requires C, 52·6; H, 7·1; N, 12·3%. Found for material dried at 100° in a vacuum : C, 56·9; H, 6·9; N, 13·0. $C_{10}H_{14}O_3N_2$ requires C, 57·1; H, 6·7; N, 13·3%). The substance recrystallised from toluene or chloroform also contains some water of crystallisation. It dissolves in chloroform to give a pure blue solution and is readily soluble in cold N/10-sulphuric acid, giving a deep blue-violet solution. It is insoluble in cold 2N-sodium hydroxide. On hydrolysis with boiling 10N-sulphuric acid it gives spinulosin in good yield (Anslow and Raistrick, *Biochem. J.*, 1938, **32**, 803). This methylamino-derivative is probably 5 : 6-*bismethylamino*-4-*methoxy*-2 : 3-*toluquinone* (see p. 1449).

(XIX) 6-Hydroxy-3-methoxy-2 : 5-toluquinone (for method of preparation, see below) (0·2 g.) was dissolved in warm ethanol (4 ml.), and alcoholic methylamine (1 ml.) added. The solution, which immediately became intensely purple, quickly deposited small purple-brown needles (0·2 g.) of 3-*methylamino*-6-*hydroxy*-2 : 5-*toluquinone*. The substance, sublimed in a high vacuum at 100—110°, formed a dark purple-red, micro-crystalline sublimate, which began to fume from 220° and melted at 252—254° (Found : C, 57·7; H, 5·2; N, 8·1; OMe, nil. $C_8H_9O_3N$ requires C, 57·5; H, 5·4; N, 8·4%). The substance is almost insoluble in cold chloroform and in water. It is readily soluble in cold 2N-sulphuric acid, giving a crimson solution, and in cold N-sodium hydroxide, giving a purple-red solution. With aqueous ferric chloride in ethanol it gives an intense purple-brown colour. On hydrolysis with boiling 2N-sodium hydroxide it gives 3 : 6-dihydroxy-2 : 5-toluquinone (Fichter, *Annalen*, 1908, **361**, 400), identified by its m. p. 184° alone or in admixture with an authentic specimen, and by its colour reactions, *i.e.*, a purple colour with cold concentrated sulphuric acid or 2N-sodium hydroxide and a dark brown colour with ferric chloride in ethanol.

6-*Hydroxy*-3-*methoxytoluquinone*. 3-Methoxy-2 : 5-toluquinone (Henrich and Nachtigall, *Ber.*, 1903, **36**, 899) (3·09 g.) was dissolved with occasional shaking in 30 ml. of a mixture of acetic anhydride (38 ml.) and concentrated sulphuric acid (2 ml.). The solution, initially blood-red, finally became pale orange and after 1½ hours colourless crystals began to separate. After standing overnight, the crystals were separated, washed with acetic anhydride, and dried (3·92 g.). The filtrate and washings were poured into ice-water; a further 1·42 g. then separated. This crude 2 : 5 : 6-*triacetoxy*-3-*methoxytoluene* crystallised from aqueous methanol in colourless hexagonal plates, m. p. 155° (Found : C, 56·7; H, 5·5; OMe, 10·8. $C_{14}H_{16}O_7$ requires C, 56·7; H, 5·5; OMe, 10·5%).

The triacetate (2 g.) was refluxed for ¾ hour with methanol (15 ml.) and concentrated sulphuric acid (0·5 ml.) in an atmosphere of nitrogen. Water was added, and the methanol removed by distillation in a vacuum. The quinol resulting from the hydrolysis was extracted with ether, the ether removed, and the crude quinol dissolved in 200 ml. of p_H 8·0 buffer solution (for composition see Section XIV). The solution, which quickly became deep purple, was aerated vigorously for 2 hours, filtered, and acidified with concentrated hydrochloric acid. 6-*Hydroxy*-

3-*methoxytoluquinone* separated in practically pure condition in orange-yellow needles (0·83 g.) and a further 0·12 g. was obtained from the filtrate by extraction with ether. It formed lustrous golden-yellow needles from light petroleum (b. p. 60—80°), m. p. 155—156° (Found: C, 56·9; H, 4·8; OMe, 18·4. $C_8H_8O_4$ requires C, 57·1; H, 4·8; OMe, 18·5%). The quinone gives a reddish-violet colour with cold concentrated sulphuric acid, a deep crimson colour immediately with 2N-sodium hydroxide, which changes quickly to a reddish-violet, and a purple colour in ethanol with aqueous ferric chloride. On methylation by the method given by Anslow, Ashley, and Raistrick (J., 1938, 439) for the methylation of 3 : 6-dihydroxytoluquinone, 6-hydroxy-3-methoxytoluquinone gives 3 : 6-dimethoxytoluquinone. The crude methylated 6-hydroxy-3-methoxytoluquinone was purified by shaking its ethereal solution with p_H 8·0 buffer solution to remove partly methylated material and sublimation in a high vacuum at 85° of the crystalline residue obtained on evaporation of the ether. Prepared in this way, 3 : 6-dimethoxytoluquinone melted at 112° (Anslow, Ashley, and Raistrick, *loc. cit.*, give 104—105°) and did not depress the m. p. of a specimen made from 3 : 6-dihydroxytoluquinone.

2 : 5 : 6-*Trihydroxy*-3-*methoxytoluene*. 6-Hydroxy-3-methoxytoluquinone (0·2 g.), shaken with a solution of sodium hyposulphite (2 g.) in water (10 ml.), dissolved almost immediately, giving a colourless solution, which was extracted with ether. The residue from the dried extract crystallised and was sublimed in a high vacuum at 90°, yielding colourless irregular prisms, m. p. 102—103°, which rapidly darkened in air to a brown and later to a purple colour (Found: C, 56·5; H, 5·8; OMe, 18·3. $C_8H_{10}O_4$ requires C, 56·5; H, 5·9; OMe, 18·2%). The *quinol* gives with cold concentrated sulphuric acid first an orange-yellow colour, which rapidly becomes apple-green, and after ¼ hour a dark emerald-green, with 2N-sodium hydroxide a deep purple colour, and with ferric chloride in aqueous or alcoholic solution an olive-brown colour which changes with excess of ferric chloride to purple.

6-*Acetoxy*-3-*methoxytoluquinone*. 6-Hydroxy-3-methoxytoluquinone does not appear to undergo the usual Thiele–Winter reaction. Instead, the hydroxy-group is acetylated and 6-*acetoxy*-3-*methoxytoluquinone* is formed. 6-Hydroxy-3-methoxytoluquinone (0·1 g.) was dissolved in 1 ml. of a mixture of acetic anhydride (19 ml.) and concentrated sulphuric acid (1 ml.), kept for 2 days, and poured into ice-water. There was a quick separation of yellow needles (0·06 g.), which were sublimed in a high vacuum at 90°, giving yellow prisms, m. p. 109° (Found: C, 57·2; H, 4·7; OMe, 14·8. $C_{10}H_{10}O_5$ requires C, 57·1; H, 4·8; OMe, 14·8%)

(XX) 4-Hydroxy-6-methoxy-2 : 5-toluquinone (Anslow, Ashley, and Raistrick, *loc. cit.*) (0·2 g.) was dissolved in cold ethanol (8 ml.), and alcoholic methylamine (1 ml.) added. The solution immediately became reddish-purple, but no solid separated even after 4 days. The solution was evaporated to dryness over concentrated sulphuric acid in a vacuum desiccator and gave a brownish-black varnish, very soluble in alcohol, from which nothing crystalline could be isolated.

(XXI) 3 : 6-Dihydroxy-4-methoxy-2 : 5-toluquinone, *i.e.*, spinulosin (Birkinshaw and Raistrick, *Phil. Trans.*, 1931, *B*, **220**, 245; Anslow and Raistrick, *Biochem. J.*, 1938, **32**, 803) (0·1 g.) was dissolved in boiling ethanol (2 ml.), and alcoholic methylamine (0·5 ml.) added. A violet product immediately separated. A further 0·5 ml. of methylamine solution was added, and the mixture kept at room temperature for several days. No further change was observed. The violet product—the *bismethylamine* salt of *spinulosin* was separated, washed with ethanol, and dried; m. p. 173° after decomposition from 164°; yield, 0·13 g. (Found: C, 48·8; H, 7·4; N, 10·8. $C_8H_8O_5,2NH_2Me$ requires C, 48·8; H, 7·4; N, 11·4%). It was insoluble in chloroform, but readily soluble in cold water to give a clear violet solution, and was immediately decomposed by cold 2N-sulphuric acid with the formation of spinulosin.

(XXII) 6-Hydroxy-3 : 4-dimethoxytoluquinone (for method of preparation, see below) (0·2 g.) was dissolved in ethanol (2 ml.), and alcoholic methylamine (1 ml.) added. The intensely purple solution quickly deposited purple-black plates, which were filtered off, washed, and dried (0·19 g.). The substance was sublimed in a high vacuum at 90—100° and crystallised first from ethanol and then from toluene, giving purple-black rectangular plates of 3-*methylamino*-6-*hydroxy*-4-*methoxy*-2 : 5-*toluquinone*, m. p. 212—213°, depressed by 10—15° on admixture with 6-methylamino-3-hydroxy-4-methoxy-2 : 5-toluquinone (see section XVII) (Found: C, 55·2; H, 5·8; N, 7·6; OMe, 15·9. $C_9H_{11}O_4N$ requires C, 54·8; H, 5·6; N, 7·1; OMe, 15·7%). The substance gives a purple (permanganate) solution in chloroform. It is readily soluble in cold N/10-sodium hydroxide to give a deep violet solution, and an alcoholic solution gives an intense olive-green colour with aqueous ferric chloride. It is not decomposed or dissolved by cold 2N-sulphuric acid and is almost insoluble in cold 5N-sulphuric acid. Hydrolysis for 3—4

minutes with boiling 2N-sulphuric acid gives, in good yield, spinulosin, identified by its m. p. 201°, alone or in admixture with an authentic specimen, and by its colour reactions.

6-*Hydroxy*-3 : 4-*dimethoxy*-2 : 5-*toluquinone*. Crude 3 : 4-dimethoxytoluquinone (2·76 g. from 8·55 g. of 5-aminohomoveratrole; Anslow, Ashley, and Raistrick, J., 1938, 441) was added to 10 ml. of a mixture of acetic anhydride (100 ml.) and concentrated sulphuric acid (5 ml.). After 24 hours it was poured into ice and water; the oil obtained did not crystallise. It was washed with water and dissolved in ether, the solution dried, and the solvent removed, giving an uncrystallisable oil (3·4 g.). (In a second experiment with pure 3 : 4-dimethoxytoluquinone, the acetylation product was sublimed in a high vacuum at 120° as a colourless oil which could not be obtained crystalline.) The crude acetylation product was refluxed for ¾ hour with 20 ml. of a mixture of methanol (60 ml.) and concentrated sulphuric acid (2 ml.) in an atmosphere of nitrogen. Water was added, and the methanol removed in a vacuum. The resulting 2 : 5 : 6-trihydroxy-3 : 4-dimethoxytoluene was extracted with ether, the solvent removed, and the residual crude phenol dissolved in 200 ml. of p_H 8·0 buffer solution. This solution, vigorously aerated for 1½ hours, quickly became deep purple. It was filtered, acidified with concentrated hydrochloric acid, and extracted with ether. On removal of the solvent the crude quinone (1·65 g.) quickly crystallised. After sublimation in a high vacuum at 65—70° and crystallisation from light petroleum (b. p. 60—80°) 6-*hydroxy*-3 : 4-*dimethoxytoluquinone* formed sheaves of tomato-red needles, m. p. 105°. It gives a blue-green colour with concentrated sulphuric acid, a purple colour with 2N-sodium hydroxide which fades on standing, and a brownish-purple colour with ferric chloride in ethanol (Found : C, 54·4; H, 5·2; OMe, 31·4. $C_9H_{10}O_5$ requires C, 54·5; H, 5·1; 2OMe, 31·3%).

2 : 5 : 6-*Trihydroxy*-3 : 4-*dimethoxytoluene*. The above quinone (0·2 g.), shaken with a solution of sodium hyposulphite (2 g.) in water (10 ml.), quickly dissolved to give an almost colourless solution, which was extracted with ether. On removal of the solvent an oil (0·2 g.), which quickly crystallised, was obtained; on sublimation in a high vacuum at 80—85° it gave colourless prisms, m. p. 110—111°. The *quinol* gives a golden-yellow colour, changing on standing to deep emerald-green, with concentrated sulphuric acid, a transitory olive-green colour, becoming finally intense purple, with 2N-sodium hydroxide, and a brownish-purple colour with ferric chloride in water or ethanol (Found : C, 54·1; H, 6·1; OMe, 31·1. $C_9H_{12}O_5$ requires C, 54·0; H, 6·0; 2OMe, 31·0%).

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